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1. CLARK*, G. E., ANDERSON, D. M. and REESE, W. V. - "Quantitative Evaluation of the Effect of Dental Pulp Irritants in Rabbit Skin and on Cultured Cells." (Abstract #350)

2. GALICH*, J. W., ANDERSON, D. M., LANGELAND, K. and REESE, W. V. - "The Treatment of Deep Carious Lesions in Naval Recruits." (Abstract #853)

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5. LEONARD*, E. P., HORTON, A. J. and MANDEL, E. J. - "The Effect of 2% Chlorhexidine Gluconate Application on Plaque and Alveolar Bone Loss in the Rice Rat." (Abstract #774)

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8. SIMONSON*, L., JACKOLA, D., LAMBERTS, B. and LEONARD, E. - "Comparison of Some Dextranase Preparations for Prevention of Dental Caries and Plaque in Hamsters." (Abstract #974)

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11. YEAGER*, J. E. and RIVIERE, G. R. - "Tooth Transplantation in RhL-A Typed Monkeys - 18 Month Postoperative Findings." (Abstract #127)

*Author presenting paper.

Quantitative Evaluation of the Effect of Dental Pulp Irritants in Rabbit Skin and on Cultured Cells. **G. E. CLARK*, D. M. ANDERSON and W. V. REESE. Naval Dental Research Institute, Great Lakes, IL**

This study was conducted to compare two methods for measuring amounts of pulp irritants in human dental caries. Irritants present in carious dentin were quantitatively evaluated for their effect on vascular permeability in rabbit skin and release of chromium (^{51}Cr) from cultured cells. Carious dentin was obtained by excavation of deep lesions under aseptic conditions. The carious material was homogenized in phosphate buffered saline (PBS), centrifuged, filtered through a 0.22 µm filter, concentrated by a Diaflo UM-2 membrane, dialized vs. deionized water, and freeze-dried. Sound dentin extract from unerupted 3rd molars was prepared in the same manner. Rabbits were prepared for the skin test by I.V. injection of Nembutal and Evans Blue dye. Intradermal injections of 0.2 ml of carious and sound dentin extracts in PBS were then made into the depilated dorsum. As little as 2 µg of caries extract resulted in a dark blue wheal within 15 min. Up to 20 µg sound dentin extract caused no wheal reaction up to 1 hr. post-injection. Both carious and sound dentin extracts effected release of ^{51}Cr from KB cells during the chromium release test (Spangberg, 1973). A minimum of 5.0 mg carious extract was required to cause a greater release of ^{51}Cr from the cells than a like amount of sound dentin extract. The rabbit skin test required considerably less material to show the presence of irritants in human carious dentin than the chromium release test.

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The Treatment of Deep Carious Lesions in Naval Recruits. J. W. GALICH*, D. M. ANDERSON, K. LANGELAND, and W. V. REESE. Naval Dental Research Institute, Great Lakes, Illinois

Pulp disease remains the most common etiologic factor in dental emergency treatments. To determine the effectiveness with which deep carious lesions are managed, incoming male recruits were screened for deep caries which involved more than three-fourths of dentin thickness, but did not appear as pulp exposures on radiographs ("U" lesions). After an evaluation of pulp health, the materials and techniques used in treatment and the observations of the operative clinician were recorded. Of 590 men screened, 86 presented a total of 134 teeth with "U" lesions. Data regarding subsequent treatment was available for 102 teeth. It indicated that 64% were excavated thoroughly, with no clinical exposure of the dental pulp and were restored. Another 4% had pulp exposures at excavation and received direct pulp capping treatment prior to restoration. Fifteen percent were treated with indirect pulp capping procedures to avoid pulp exposure. Root canal fillings were placed in 8% of the teeth and less than 10% were extracted, after being judged unmanageable. Short term follow-up examinations (ranging from 2 to 12 months) suggested a high degree of success since none have required retreatment or extraction. Thus more than 82% of the original 102 teeth received some form of conservative pulp treatment with the dental pulp intact. The efficacy of restoring pulp health by these various types of treatment continues to be monitored through long-term follow-up examinations.

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Histologic Assessment of Periodontal Probes in Normal Gingiva.
E. B. HANCOCK*, M. R. WIRTHLIN and J. ELLINGSON. Naval Dental Research Institute, Great Lakes, Illinois

There has been some question for a number of years as to what clinical probing measurements indicate. The purpose of this study was to evaluate the use of the periodontal probe as a non-invasive method for assessment of attachment level in normal gingiva. Amalgam restorations were placed on the labial surfaces of the right central incisors in 14 adult Rhesus monkeys (mean age=65.3 mo.). Gold-coated acrylic replicas of periodontal probes with Williams' markings were placed in the crevice with a light probing pressure, and luted to the tooth with cyanoacrylate. Clinical measurements were made from the amalgam preparation to the gingival margin and to the base of the crevice. The gingiva appeared clinically healthy in all animals and the crevices were shallow (1.6±.47mm). The tooth, probe gingiva, and alveolar crest were immediately removed in block section, decalcified, and processed histologically. Evaluations were possible in 10 specimens. Histologically, there were scattered inflammatory cells underlying the epithelium. In all specimens the tip of the probe was coronal to the CEJ (3.0±.55mm); in 6 the tip ended in the area of the junctional epithelium; in 4 specimens the tip of the probe was at the transition from crevicular to junctional epithelium. At the tip of the probe there was compression of the junctional epithelium and connective tissue. In normal gingiva the tip of the probe rested within epithelium at or slightly apical to the coronal extent of the junctional epithelium. Thus probe measurements in normal gingiva would indicate the base of the crevice but not the level of the connective tissue attachment.

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Glucanase-Producing Organisms in Dental Plaque of Caries-Free and Caries-Active Naval Recruits. B. L. LAMBERTS*, I. L. SHKLAIR, R. G. WALTER, and S. SHELTON. Naval Dental Research Institute, Great Lakes, Illinois

The aim of this study was to compare dental plaque samples of caries-free and caries-active naval recruits for the prevalence of glucanase-producing organisms. Plaque samples were collected from 15 caries-free and 14 caries-active subjects by flossing two posterior interdental contacts in each quadrant. The two samples from each quadrant were combined in thioglycolate broth holding medium, sonicated, diluted, and transferred to heart-infusion agar plates which contained 0.1% of either one of two glucans as substrates to detect the glucanases. One type of plate contained blue dextran to locate dextranases (Staat et al., 1973); the other type contained the nonsolubilized products from dextranase-degraded water-insoluble glucans of S. mutans strains K-1R or OMZ 176, in an effort to detect α-1, 3 glucanases. The plates were incubated at 37 C for 5-7 days in 5% CO_2-95% N_2 atmosphere, and total and glucanase-producing colonies were counted. Although dextranase-producing colonies (d.p.c.) were evident in all subjects, no α-1, 3 glucanase-producing organisms were found. The d.p.c. percentages according to dental quadrant showed broad within-subject variation, but there were no significant differences among the quadrant means within each group. Mean d.p.c. percentages below 2.0 were found for 10 of the 15 caries-free subjects, but for only 4 of the 14 caries-active subjects. However, the overall d.p.c. percentages for the caries-free subjects (mean ± s.d. = 3.60 ± 4.98) were comparable to those of the caries-active subjects (mean ± s.d. = 3.83 ± 5.69).

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The Effect of 2% Chlorhexidine Gluconate Application on Plaque and Alveolar Bone Loss in the Rice Rat. **E. P. LEONARD*, A. J. HORTON and E. J. MANDEL. Naval Dental Research Institute, Great Lakes, Illinois**

The purpose of this study was to observe the effect of daily swabbing with 2% chlorhexidine gluconate on the accumulation of dental plaque and the loss of alveolar bone in the rice rat. Weanling rice rats (Oryzomys palustris) were placed on L-2000 diet and divided into 3 groups. One group was treated 5 times per week with topically applied 2% chlorhexidine gluconate. One control group was swabbed with distilled water in a similar manner and a second control group was not swabbed. The animals were killed at 60 $\pm$ 2 experimental days. The upper and lower jaws were dissected free and the total area of plaque accumulation was determined in all quadrants with the aid of a disclosant, a 10x dissecting scope and a reference grid. Half of each mandible was fixed, decalcified and cut in a cryostat for histologic study. Contralateral halves were defleshed and alveolar bone loss was quantitated along buccal and lingual surfaces by direct measurement with a micrometer eyepiece. Comparison of values were made by Welch's t test. The results of analysis showed a significant reduction in plaque in the chlorhexidine group when compared to either of the control groups ($p<0.001$). Alveolar bone scores revealed significantly more bone loss ($p<0.001$) in the control groups than in the chlorhexidine recipient group. Additionally a comparison of control groups revealed significantly more bone loss ($p<0.001$) in the unswabbed group. The results indicated that the topical application of 2% chlorhexidine gluconate decreases the amount of plaque accumulation and the rate of alveolar bone loss in this animal model.

Supported by NMR&DC Project No. MR04120.MR0412002.0408.

An Evaluation of Indices in the Classification of Naval Recruits into High and Low Risk Caries Groups. **E. MANDEL*, M. R. WIRTHLIN and R. G. WALTER. Naval Dental Research Institute, Great Lakes, Illinois**

A group of 130 naval recruits was evaluated in an attempt to identify high and low risk caries patients by the use of caries and periodontal indices, and demographic variables. Each recruit was examined upon entering the Navy and again 6 months later. Each recruit was then classified by the severity of his carious lesions at the first examination as follows: Low risk group - at least one tooth with a lesion extending 1/4 to 1/2 into the dentin (N=67); and High risk group - at least one tooth with a lesion extending more than 1/2 into the dentin; lesions of lesser depth could also exist (N=49). Fourteen recruits had no detectable lesions at the first exam and were not considered further. The mean of each index in the high risk group was compared by t-test to the mean of the same index in the low risk group. For both examinations in the high risk group, the means of DMFT, DMFS, missing surfaces, and Calculus Surface Index were significantly greater (p between 0.02 and 0.001); surfaces-at-risk (SAR) and posterior proximal surfaces-at-risk (PPSAR) were significantly lower ($p<0.001$). In addition, means for the caries attack rates (whole mouth and posterior proximal only) were significantly greater ($p<0.05$). None of the other periodontal or demographic indices showed such significance. These results suggested that DMFT, DMFS, SAR and PPSAR could be used in the classification of patients into high and low risk caries groups. The caries attack rates appeared to validate the identification of the caries risk groups, and suggested using depth of lesions as an additional criterion for classification.

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S. mutans Glucan Production and Caries Activity in Rats. I. L. SHKLAIR*, R. G. WALTER, E. P. LEONARD and S. SHELTON. Naval Dental Research Institute, Great Lakes, Illinois

The purpose of this study was to determine the amount of extracellular glucan produced by each of 5 *S. mutans* serotype c strains and assess its relationship to caries activity in animals. The organisms were grown in 5 ml of a chemically defined medium with 5% sucrose and the amount of soluble and insoluble glucans (expressed as mg/ml of glucose equivalents) were determined before and after animal implantation. The organisms were implanted into weanling antibiotic treated rats; the animals were then maintained on the cariogenic diet 2000. The animals were killed 60 days after implantation and their caries scores determined by the procedure of Keyes. Strain 1 was classified as a low glucan producer, 1.20 mg/ml of glucose equivalents. It produced nearly equal amounts of soluble and insoluble glucans. This organism caused a proximal caries score of 4.4 in the rats. Strain 2 produced 2.05 mg/ml of glucans. Glucan production of this recovered organism at sacrifice rose to 2.65 mg/ml; all of this glucan was insoluble. The proximal caries score produced by this organism was 11.5. Strains 3 and 4 produced glucan levels of 3.26 and 3.55 mg/ml, with proximal caries scores of 7.1 and 15.4 respectively. Approximately 85% of the glucans produced by both organisms were insoluble. Strain 5 produced 6.0 mg/ml of glucans; however, only about 1/3 was insoluble. Strain 5 produced a proximal caries score of 14.75. A trend is noted from the data, that the amount of glucans produced by S. mutans, usually insoluble glucans, could be related to proximal caries activity in rats.

Supported by NMR&DC Project No. MR0412002.6049.

Comparison of Some Dextranase Preparations for Prevention of Dental Caries and Plaque in Hamsters. **L. SIMONSON*, D. JACKOLA, B. LAMBERTS and E. LEONARD. Naval Dental Research Institute, Great Lakes, IL**

Four dextranase preparations were compared for their oral therapeutic properties in antibiotic suppressed syrian hamsters. Six experimental groups, 10 animals/group, were orally infected with Strep. mutans NCTC 10449 (made resistant to streptomycin), fed cariogenic diet 2000, and killed 80 days after implantation. Group 1 consisted of uninfected controls; Group 2, the positive controls, received no therapeutics. Groups 3 and 4 received dextranase derived from Fusarium moniliforme (FM) and Penicillium sp. respectively ad libitum in their drinking water (100 units/ml). Group 5 was swabbed 2-3 times per week with a preparation of FM-dextranase (80 units/ml) which was chemically altered to increase its affinity for enamel. Group 6 was treated similar to Group 5, except unaltered FM-dextranase was applied. No significant weight gain difference was noted between the groups 21 and 80 days after implantation ($p \leq 0.05$). Quantitative data on total plaque, lingual and buccal bone-loss amoung the groups were not significantly different ($p \leq 0.05$). However, when total caries scores between the groups were compared Group 3, and to a lesser extent Group 4, had significantly lower caries scores ($p < 0.05$) than the positive control group. Although Group 6 had less total caries than Group 2 (positive control) it was not a significant difference ($p \leq 0.05$). Unexpectedly, Group 5 had significantly elevated total caries scores ($p < 0.05$) relative to Group 2. The data indicated that although dextranase can potentially lower caries scores, its topical application on unprepared enamel was not effective.

Supported by NMR&DC Project No. MF51524012.0021.

A 20 Year Comparison of Disease Experience and Treatment Requirements in Naval Recruits. R. G. WALTER*, M. R. WIRTHLIN, and E. B. HANCOCK. Naval Dental Research Institute, Great Lakes, Illinois

This study had two purposes: (1) to relate changes in the disease experience of naval recruits during the last 20 years and (2) to determine the current initial, accomplished and remaining treatment requirements during their first six months of naval training. A randomly selected group (N=161) of recruits and a group preselected for service school began the study. The dental caries examination was conducted by one investigator with the aid of radiographs. The periodontal disease status of all subjects was evaluated by calibrated investigators. A treatment plan was formulated for each subject based on criteria developed by senior clinicians. The caries indices used in this study included DMFT and DMFS. Because the caries indices of the random group and the selected group were not statistically different, they were combined into a single cluster of 397 subjects. Statistical comparisons (using Welch's "t" test) of the present data with data collected in a similar study in 1956 demonstrated a statistically significant reduction ($p<0.001$) in the mean number of DT (5.3 vs 7.0) and DS (7.8 vs 12.5). No other comparisons of caries indices proved statistically significant. Treatment requirements per 1000 recruits included the following procedures: 13,700 operative and prosthodontic, 500 oral surgery, 8,500 periodontic and oral hygiene, and 6,100 radiographic and miscellaneous. During the subjects first six months of service, 63% of the required dentistry was completed. However, 37% of the initial requirements remained and an additional 13% was required.

Supported by NMRDC Project No. ZF51524012.0006.

Clinical, Epidemiological, and Behavioral Profiles of Young Adult Males with Necrotizing Gingivitis. M. R. WIRTHLIN*, and L. DEVINE.
Naval Dental Research Institute, Great Lakes, Illinois

The high incidence of necrotizing gingivitis in young adult servicemen is unusual for a disease attributed to fatigue, disability, lowered resistance, tobacco smoking and emotional stress. The purpose of the investigation was to seek a case history pattern which could account for a high incidence in a segment of the population which is usually in good health. Data from clinical examination, laboratory tests, history, environment, diet, mental state, interpersonal contacts, and microbiological samples were collected on 15 subjects. The subjects were all white, male, unmarried, 17 to 25 years of age, of low socioeconomic background with high school or less education. They were in the lower military paygrades and in the first two years of service. There were complaints of malaise, upper respiratory illness symptoms but no common medical condition. Dental findings were not markedly different from comparable servicemen except for the necrotizing gingivitis. A two-day diet recall indicated a nutritionally adequate diet. Subjects generally denied there was stress associated with dental visits, environment, diet, or duties. Half of the subjects related subjective or objective findings of mental or emotional stress. Six had self-rating depression scale indices (Zung 1965) under 40 (normal), 2 had intermediate scores, and 7 had over 50 reflecting possible emotional disorders and depression. Promiscuous sexual behavior was a common finding. A contact was reported by 14 subjects, usually about one to four weeks before onset of symptoms. Preliminary microbiological samplings from the necrotic lesions were negative for the isolation of incriminating organisms.

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Tooth Transplantation in RhL-A Typed Monkeys - 18 Month Postoperative Findings. J. E. YEAGER*, and G. R. RIVIERE. Naval Dental Research Institute, Great Lakes, IL and UCLA, California

The objective of this study was to determine the short and long term responses of genetically matched Rhesus monkeys to dental allografts. Thirty animals ranging in age from 25 to 65 months were selected for this project. Each monkey received one autologous and one allogeneic bicuspid tooth transplant. All donor teeth that had erupted at the time of surgery were placed in erupted positions; unerupted donor teeth were transplanted into positions completely within alveolar bone and were covered with mucoperiosteal flaps. Radiographic, clinical, histopathological and immunological findings were used in evaluating recipient responses. Since one monkey died 7 months postoperatively, 29 animals were included in the long term evaluation. At 18 months, 22 of 29 allografts and 28 of 29 autografts remained. Submersion and resorption were common among the allogeneic and infrequent among the autologous transplants. Although 5 allografts and 22 autografts were determined to be at least partially functional 18 months postoperatively, none of the allogeneic and only 17 of the autologous transplants were both fully erupted and firm. At least one allograft was exfoliated from animals in each of the 5 groups in which the recipients were of varying degrees of RhL-A disparity (N=8,4,2,1,1). The allogeneic control group, which consisted of monkeys that were completely RhL-A donor/recipient disparate (N=10), lost 2 allografts. The allogeneic transplants in the RhL-A completely matched groups (N=3) were retained for the entire study period, but were clinically inferior to their autologous counterparts.

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